In The Name of God

Ardabil University of Medical Sciences

School of Medicine

Submitted to the School of Medicine in Partial Fulfillment of the Requirements for the Degree of Master of Science (M.Sc.) in Clinical Biochemistry

Subject:

The Effect of Dimethyl Sulfone on Gastrointestinal Cancer Cell Lines (AGS, HepG2 and KYSE-30) and Flow Cytometric Analysis of the Cell Cycle

Supervisor:

Dr. Shahab Bohlooli

Advisor:

Dr. Mohammad Mazani

By: **Naser Jafari**

Dedication

Dedicated to:

My Unfailing Kind ***Mother***

Who has been supportive of me in every aspect of life,

And to My Brother and Sister:

Saeed and ***Mahdieh***

For their kindness

And

To my kind professor ***Dr. Bohlooli,***

A brilliant researcher, scientist, and teacher,
For training me to become a researcher, I have learned so much in the lab, it is impossible to recount the details in this space.
Thanks for all of your help and advice, and for providing me with so many opportunities to discover.

To researchers and scientists, who with their hard work pave the road to discovery,

Last but not least, to anyone who has been touched by cancer, as they are my motivation for this thesis and for my research.

Acknowledgments

My sincere thanks are due to the friends and people, who made my research possible,

Including gentlemen: Khanzadeh, Hosseini, Rostami, Mohammadnia, Khodayi, Khaligh, Shahbazzadegan,

And ladies: Naftchi, Golmaghani, Esmaeeli, Molazadeh and Mohandesi

Thank you all

I am grateful of my friends, who made my life outside of lab exciting,

Saeed Aslani, Poorya Badihi, Aboozar Barzegar, Rahim Delsouzi, Nemat Heydarogloo, Naser Jafargholizadeh, Yaser Khakpour, Amir Mahmoodzadeh, Yashar Nabizadeh, Mehrdad Nickmanesh, Rezgar Rahbari, Mehrdad Sarvghamat, Ali Shadmanfar, Hasan Shaghaghi, Saeed Zarenejad.

Thanks for everything,

Abstract:

Background: Methylsulfonylmethane (MSM) is a dietary supplement to support healthy body. It is considered to be one of the least toxic substances in biology. Considering preventive effects of this substance on tumor onset and its low toxicity to healthy body we investigated in vitro cytotoxic effects of methylsulfonylmethane on cancer cell lines. **Methods**: AGS, HepG2 and KYSE-30 cancer cell lines were treated by MSM by initial concentration of 50 mg/ml with ¼ serial dilutions and incubated for 24, 48 and 72 hours. Cytotoxicity was examined through MTT, neutral red uptake and protein measurement assays. EB/AO staining was used for apoptotic cell detection. A DAPI staining method was used to analysis cell cycle by flow cytometry. **Results**: IC50 of the MSM on AGS, HepG2 and KYSE-30 cell lines were 28.04, 21.87 and 27.98 mg/ml after 72 hours respectively. The EB/AO staining showed an increase in apoptotic cells after MSM treatment. The flow cytometry cell cycle analysis showed a significant increase in cell density at G2/M phase. **Conclusion**: MSM had cytotoxic effect on cancer cell lines but HepG2 cell line was more susceptible. This study provides the evidence that MSM may induce cytotoxic effect on cancer cell lines by apoptosis and it could be of value in realm of chemotherapy.

Key Words: Methylsulfonylmethane, MSM, Cancer cell lines, Cytotoxicity, Apoptosis, Adjuvant

Abbreviations:

Abbreviation	Text
°C	Centigrade
µg	Micro-gram
µl	Micro-liter
AO	Acridine Orange
BBB	Blood Brain Barrier
CAT	Catalase
cm	Centimeter
COX	Cyclooxygenase
DAPI	4',6'-Diamidino-2-Phenylindole
DMSO	Dimethyl Sulfoxide
$DMSO_2$	Dimethyl Sulfone
DNA	Deoxyribonucleic Acid
EB	Ethidium Bromide
FBS	Fetal Bovine Serum
FDA	Food and Drug Administration
FRAME	Fund for the Replacement of Animals in Medical Experiments

g	Gram
G1	Gap 1 Phase
G2	Gap 2 Phase
GC	Gastric Cancer
GI	Gastrointestinal
GSH	Glutathione
h	Hour(s)
HBV	Hepatitis B Virus
HCC	Hepatocellular Carcinoma
HCV	Hepatitis C Virus
IC	Interstitial Cystitis
IC50	The Half Maximal Inhibitory Concentration
Ig E	Immunoglobulin E
IL-1β	Interleukin 1 Beta
IV	Intravenous
kPa	Kilopascal
kg	Kilogram
LDL	Low Density Lipoprotein
M	Mitosis

MDA	Malondialdehyde
mg	Milligram
min	Minute
ml	Milliliter
mol wt	Molecular Weight
MPO	Myeloperoxidase
MSM	Methylsulfonylmethane
MTT	MTT (3-(4,5-Dimethylthiazol-2-yl)-2,5 Diphenyltetrazolium Bromide)
NAD(P)H	Nicotinamide Adenine Dinucleotide (Phosphate)
NIH	National Institute of Health
nm	Nanometer
OD	Optical Density
PBS	Phosphate Buffered Saline
pg	Pico-gram
PGE2	Prostaglandin E2
ppm	Parts Per Million
rcf	Relative Centrifugal Force
rpm	Round Per Minute
S	Synthesis Phase

SD	Standard Deviation
SEM	Standard Error of the Mean
UV	Ultraviolet
w/v	Weight/Volume

Figures and Tables

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1: Introduction

1.1 Cancer:

Cancer causes about one-fifth of the deaths each year. About 100-350 of 100,000 people scarify by cancer in a year. Cancer is due to failure of the mechanisms that usually control the growth and proliferation of cells. Throughout adult life and during normal development, genetic control systems regulate the balance between cell birth and death in response to various signals. Cell birth and death rates determine adult body size. The cells in many adult tissues normally do not proliferate except during healing processes. Such stable cells (e.g., hepatocytes, gastric and intestinal epithelial cells, and white blood cells) can remain functional for long periods or even for the entire lifetime of an organism. Cancer occurs when the mechanisms that maintain these normal growth rates malfunction to cause excess cell division. The failure of cellular regulation that give rise to most or all cases of cancer are due to genetic damage that is often accompanied by influences of tumor-promoting chemicals, hormones and sometimes viruses [1] (Figure 1-1).